Learning and memory II
Fear conditioning: consolidation, reconsolidation and other friends

System's neurophysiology
Dana Cohen
Cellular Memory Consolidation Theory

Short-Term Memory (STM)
- Seconds to Hours
- ”Labile” (sensitive to disruption)
- Does not require new RNA & protein synthesis

Long-Term Memory (LTM)
- Days, Weeks, lifetime
- Consolidated (insensitive to disruption)
- Does require new RNA & protein synthesis
Fear Conditioning

Conditioned Stimulus (CS)
e.g. light or tone

Unconditioned Stimulus (US)
e.g. footshock

Natural Threat → Amygdala

Defensive behavior
Autonomic arousal
Hypoalgesia
Reflex potentiation
Adrenal activation
Within-subjects design (Anagnostaras et al., 1999)

REMOTE TRAINING

10 Tone-Shock Pairings
Within-subjects design

REMOTE TRAINING

RECENT TRAINING

50 days later

10 Tone-Shock Pairings
(different tone)

Which contexts and tones Used were counterbalanced
Within-subjects design

REMOTE TRAINING → 50 days later → RECENT TRAINING → next day → Lesion

Electrolytic Dorsal Hippocampus Lesion

DH

SHAM

AP

0

−2

−3

−6
Within-subjects design

REMOTE TRAINING

50 days later

RECENT TRAINING

next day

Sham or Hippocampus Lesion

10 day recovery

REMOTE CONTEXT

Test Order Counterbalanced

RECENT CONTEXT

next day
DH lesions spare remote context memory

![Graph showing freezing behavior over time]

**Remote Contextual Fear**

- **Freezing (% time)**
- **Minutes**
- **DH**
- **Sham**
DH lesions disrupt recent context memory

A. Remote Contextual Fear

B. Recent Contextual Fear

Freezing (% time)

Minutes

Sham

DH
DH lesions produce a time-limited retrograde amnesia of contextual fear.

A) Remote Contextual Fear
B) Recent Contextual Fear
C) Context Summary

Freezing (% time)

Minutes

Sham
DH
Within-subjects design

REMOTE TRAINING

50 days later

RECENT TRAINING

next day

Sham or Hippocampus Lesion

10 day recovery

REMOTE CONTEXT

next day

RECENT CONTEXT
Within-subjects design

Third Context for Tone Testing Only

- Baseline Period
- Dark Red Lighting
- Quiet
- Pine Shavings Odor
- No Grids
- Like Home Cage

Sham or Hippocampus Lesion

10 day recovery

REMOTE TRAINING

RECENT CONTEXT

REMOTE TONE
going to next day

REMOTE TONE
going to next day

REMOTE CONTEXT
going to next day

Third Context for Tone Testing Only

RECENT TONE
Within-subjects design

REMOTE TRAINING

50 days later

RECENT TRAINING

next day

Sham or Hippocampus Lesion

10 day recovery

RECENT TONE

next day

REMOTE TONE

next day

REMOTE CONTEXT

next day

RECENT CONTEXT
DH lesions spare remote tone fear memory

Freezing (% time)

Minutes

Remote Tone Fear

Sham
DH
DH lesions spare recent tone fear memory

**A** Remote Tone Fear

**B** Recent Tone Fear

<table>
<thead>
<tr>
<th>Freezing (% time)</th>
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<tbody>
<tr>
<td>100</td>
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<tr>
<td>75</td>
</tr>
<tr>
<td>50</td>
</tr>
<tr>
<td>25</td>
</tr>
<tr>
<td>0</td>
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</tbody>
</table>

![Graph A: Remote Tone Fear](image)

![Graph B: Recent Tone Fear](image)

Legend:
- **Sham**
- **DH**
DH lesions spare tone fear memory

A. Remote Tone Fear
B. Recent Tone Fear
C. Tone Summary

Freezing (% time)

Minutes

Remote (six min)

Sham

DH
DH lesions produce a highly selective deficit in recent contextual fear memory

Content could be:
- Context-shock association
- Just memory of the context
Impaired acquisition of fear conditioning

Re-exposure to the CS (tone) for 60 seconds after 24 hours

Nader et al. 2001
Inactivation of the amygdala before but not after auditory fear conditioning prevents memory formation:

Pre-training injections of muscimol

Post-training injections of muscimol

Wilensky et al. 1999
Does the Consolidation of Auditory Fear Conditioning Require Protein Synthesis in the LA?

Basic Paradigm:

1 x Tone-Shock → $4 \text{ hr}$ → STM → $20 \text{ hr}$ → LTM

Anisomycin infusion into the Lateral Amygdala

Schafe & LeDoux, 2000
Protein synthesis inhibition in the LA blocks the induction of long term memory.

1 x CS-US $\xrightarrow{4 \text{ hr}}$ STM $\xrightarrow{20 \text{ hr}}$ LTM

Schafe & LeDoux, 2000
The effect of memory retrieval

• ECS following a brief presentation of fear conditioned CS caused retrograde amnesia
• ECS alone did not cause retrograde amnesia

Possible interpretations:
- Amnesia was caused by the destruction of the memory trace
- The memory was temporarily inaccessible
Do Consolidated Memories Return to a Labile State When Retrieved or Reactivated?

Consolidation:

1 x Tone-Shock \(\rightarrow\) STM \(\rightarrow\) LTM

Anisomycin infusions into the Lateral amygdala

Reconsolidation:

1 x CS-US \(\rightarrow\) CS \(\rightarrow\) PR-STM \(\rightarrow\) PR-LTM

Nader, Schafe & LeDoux, 2000
Predictions

Tone-Shock $\xrightarrow{24\ hr}$ CS $\xrightarrow{4\ hr}$ PR-STM $\xrightarrow{20\ hr}$ PR-LTM

1- If reactivation of a consolidated memory causes it to undergo another time-dependent memory stabilization process then post-reactivation anisomycin infusions should block PR-LTM but not PR-STM.

2- If consolidated memories remain fixed in the brain, then post-reactivation anisomycin should have no detrimental effect on the memory.
Protein synthesis inhibition in the LA blocks consolidation and reconsolidation.

Consolidation

Reconsolidation

Schafe & LeDoux, 2000

Nader, Schafe & LeDoux, 2000
A Test of Whether Reconsolidation Depends on Reactivation of the Memory

1 x CS-US $\xrightarrow{24\ hr}$ No CS $\xrightarrow{24\ hr}$ Test 2
Anisomycin’s behavioral effects depend on memory reactivation

1 x CS-US $\xrightarrow{24\ hr}$ No CS $\xrightarrow{24\ hr}$ Test

![Graph showing percent freezing over three trials with error bars]

Title: Anisomycin’s behavioral effects depend on memory reactivation

Graph: A line graph showing percent freezing over three trials with error bars, indicating variability in the freezing response. The x-axis represents trial numbers (1, 2, 3), and the y-axis represents percent freezing (0 to 100).
So far

By definition;

- Given that anisomycin had no effect on the memory when the memory was not reactivated demonstrates it was in a consolidated state.

- Given that anisomycin impaired the memory when the memory was reactivated demonstrates it was in a labile state.

Therefore, the reactivation of consolidated auditory fear memories returns them to a labile protein synthesis dependent state in the LA.
Lewis' Memory Model

Active Memory
- Seconds to Hours
- "Labile" (sensitive to disruption)
- *Does require new RNA & protein synthesis*

Inactive Memory
- Days to Weeks
- Consolidated (insensitive to disruption)
- *Does not require new RNA & protein synthesis*

Lewis, 1979
Changes in Body Length
Conditioned malaise - Sea Slug

PR-STM  PR-LTM
-4 -3 -2 -1 0 1 2 3

Control
Anisomycin

(PR-STM) (PR-LTM)

(Nader et al, 2000)

Percent Freezing
Auditory fear conditioning - **Rats**
Intra-amygdala infusions

Percent Freezing
Context fear conditioning - **Rats**
Intra-hippocampus infusions

Percent Freezing
Contextual fear conditioning - **Mice**
Inducible dominant negative

Vehicle
Anisomycin

(PR-STM) (PR-LTM)

(Control)

(Debiec et al, 2002)

(Anisomycin)

(CREB I)

(Control)

(Kida et al, 2001)

Object recognition - **Mice**
Transgenic Knockout

Percent Exploration
Motor sequence learning - **Humans**

Percent Change From Reactivation

(Control)

(Zif286KO)

(Control)

(Interference)

(Bozon et al, 2003)

(Walker & Stickgold, 2003)
Memory reconsolidation is time dependent

Contextual fear conditioning: 1 foot shock

A. Injection
   CS-US \( \rightarrow \) CS
   | 2h  | 24h |
   \[ \text{Percent Freezing} \]
   \[ \text{control} \text{ ANI} \]

B. Injection
   CS-US \( \rightarrow \) No CS \( \rightarrow \) CS
   | 24h (0 min) | 24h |
   \[ \text{Percent Freezing} \]
   \[ \text{control} \text{ ANI} \]

C. Injection
   CS-US \( \rightarrow \) CS(1 min) \( \rightarrow \) CS
   | 24h  | 24h |
   \[ \text{Percent Freezing} \]
   \[ \text{control} \text{ ANI} \]

D. Injection
   CS-US \( \rightarrow \) CS(3 min) \( \rightarrow \) CS
   | 24h  | 24h |
   \[ \text{Percent Freezing} \]
   \[ \text{Re-exposure} \text{ TEST} \]

E. Injection
   CS-US \( \rightarrow \) CS(30 min) \( \rightarrow \) CS
   | 24h | 30 min | 24h |
   \[ \text{Percent Freezing} \]
   \[ \text{control} \text{ ANI} \]

Suzuki, JNS 2004
Memory reconsolidation is strength dependent

Contextual fear conditioning: 3 foot shock

Suzuki, JNS 2004
Memory reconsolidation is age dependent

Contextual fear conditioning: 1 foot shock

A 1 week (CS; 3min)    B 3 weeks (CS; 3min)

C 8 weeks (CS; 3min)    D 8 weeks (CS; 10min)

Suzuki, JNS 2004
Does reactivation of one component of a memory return associated memories to a labile state?

Debiec, Doyer, Nader & LeDoux
Second order fear conditioning generates a small associative network

- The CS (CS1) is paired with the US
- CS2 is repeatedly paired with CS1 in the absence of the US

Debiec, et al PNAS 2006
Validating SOFC by extinction

An associative network is formed in which

\[ CS2 \rightarrow CS1 \rightarrow US \]

Debiec, et al PNAS 2006
Does reactivation of one component of a memory return associated memories to a labile state?

- Direct reactivation of the first order memory causes it to undergo reconsolidation.
- When the first order memory is indirectly reactivated it does not return to a labile state.

Debiec, et al PNAS 2006
• The reconsolidation hypothesis states that reactivated memories require new protein to restabilize

• It is fundamentally different from the idea that retrieval of a memory makes it vulnerable to the effects of a subsequent event
controversy

• Could anisomycin by itself (like ECS) modify the neural network activity and thus actively cause amnesia?

• Note that anisomycin is used in cancer research to study apoptosis
Brief re-exposure inhibitory avoidance behavior

Power, Learn Mem 2006
• These findings are in conflict with the reconsolidation hypothesis, but are consistent with the alternative interpretations that such treatments may temporarily disrupt memory retrieval.

Rudy, Learn Mem 2006
• The emphasis on retrieval failure as the source of forgetting in psychobiology has been growing and making increasing inroads on the traditional storage-failure approach.

• The major lesson for psychobiologists is that they would profit from turning a greater amount of their research efforts to the study of retrieval mechanisms and that they should consider the implications for physiological study of a learning engram that is formed permanently in less than a second.

Rudy, Learn Mem 2006
Are consolidation and reconsolidation similar or distinct processes?
BDNF antisense ODN disrupts memory consolidation but not reconsolidation


- BDNF antisense ODN
- BDNF missense ODN
zif268 antisense ODN disrupts memory reconsolidation but not consolidation

Lee, et al science 2007
Erasure of Long-Term Memory Associations in the Cortex

persistent phosphorylation by PKMζ is critical for storage of long-term memory in cortex.

Application of protein kinase C inhibitor – prevents phosphorylation by PKMζ

Science, 2007, 317 (951-3)
Possible mechanisms

• Temporal window of “cellular consolidation” continues far longer than originally thought.
• PKMζ permanently maintains long-term memory